

PCT

RAW SEQUENCE LISTING DATE: 07/16/2004
PATENT APPLICATION: US/10/501,071 TIME: 16:37:40

Input Set : A:\Sequence Listing.ST25.txt
Output Set: N:\CRF4\07162004\J501071.raw

```
5 <110> APPLICANT: University of Newcastle Upon Tyne
     9 <120> TITLE OF INVENTION: Fusion Proteins
     13 <130> FILE REFERENCE: 43952/JMD/MAR
C--> 17 <140> CURRENT APPLICATION NUMBER: US/10/501,071
C--> 17 <141> CURRENT FILING DATE: 2004-07-09
     17 <150> PRIOR APPLICATION NUMBER: GB 0200689.8
     19 <151> PRIOR FILING DATE: 2002-01-10
     23 <160> NUMBER OF SEQ ID NOS: 61
     27 <170> SOFTWARE: PatentIn version 3.1
     31 <210> SEQ ID NO: 1
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     35 <212> TYPE: PRT
     37 <213> ORGANISM: Artificial Sequence
     41 <220> FEATURE:
     43 <223 > OTHER INFORMATION: Ala3-His6 tail
     45 <400> SEQUENCE: 1
     47 Ala Ala Ala His His His His His
     51 <210> SEQ ID NO: 2
     53 <211> LENGTH: 25
     55 <212> TYPE: PRT
     57 <213> ORGANISM: Escherichia coli
     61 <400> SEQUENCE: 2
     63 Met Asn Met Lys Lys Leu Ala Thr Leu Val Ser Ala Val Ala Leu Ser
                      5
     67 Ala Thr Val Ser Ala Asn Ala Met Ala
                  20
     71 <210> SEQ ID NO: 3
     73 <211> LENGTH: 5
     75 <212> TYPE: PRT
     77 <213> ORGANISM: Artificial Sequence
     81 <220> FEATURE:
     83 <223> OTHER INFORMATION: Cleavage site for enterokinase
     85 <400> SEQUENCE: 3
     87 Asp Asp Asp Lys
     91 <210> SEQ ID NO: 4
     93 <211> LENGTH: 4
     95 <212> TYPE: PRT
     97 <213> ORGANISM: Artificial Sequence
     101 <220> FEATURE:
     103 <223> OTHER INFORMATION: Cleavage site for thrombin.
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105 <400> SEQUENCE: 4

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107 Leu Val Pro Arg 108 1 111 <210> SEQ ID NO: 5 113 <211> LENGTH: 4 115 <212> TYPE: PRT 117 <213> ORGANISM: Artificial Sequence 121 <220> FEATURE: 123 <223> OTHER INFORMATION: Cleavage site for factor Xa 125 <400> SEQUENCE: 5 127 Ile Glu Gly Arg 128 1 131 <210> SEQ ID NO: 6 133 <211> LENGTH: 4 135 <212> TYPE: PRT 137 <213> ORGANISM: Artificial Sequence 141 <220> FEATURE: 143 <223> OTHER INFORMATION: 4xHis tag 145 <400> SEOUENCE: 6 147 His His His His 148 1 151 <210> SEQ ID NO: 7 153 <211> LENGTH: 5 155 <212> TYPE: PRT 157 <213> ORGANISM: Artificial Sequence 161 <220> FEATURE: 163 <223> OTHER INFORMATION: 5xHis tag 165 <400> SEQUENCE: 7 167 His His His His 168 1 171 <210> SEQ ID NO: 8 173 <211> LENGTH: 6 175 <212> TYPE: PRT 177 <213> ORGANISM: Artificial Sequence 181 <220> FEATURE: 183 <223> OTHER INFORMATION: 6xHis tag 185 <400> SEQUENCE: 8 187 His His His His His 188 1 191 <210> SEQ ID NO: 9 193 <211> LENGTH: 7 195 <212> TYPE: PRT 197 <213> ORGANISM: Artificial Sequence 201 <220> FEATURE: 203 <223> OTHER INFORMATION: 7xHis tag 205 <400> SEQUENCE: 9 207 His His His His His His 208 1 211 <210> SEQ ID NO: 10 213 <211> LENGTH: 8

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217 <213> ORGANISM: Artificial Sequence
221 <220> FEATURE:
223 <223> OTHER INFORMATION: 8xHis tag
225 <400> SEQUENCE: 10
227 His His His His His His His
228 1
231 <210> SEQ ID NO: 11
233 <211> LENGTH: 9
235 <212> TYPE: PRT
237 <213> ORGANISM: Artificial Sequence
241 <220> FEATURE:
243 <223> OTHER INFORMATION: 9xHis tag
245 <400> SEQUENCE: 11
247 His His His His His His His His
248 1
251 <210> SEQ ID NO: 12
253 <211> LENGTH: 10
255 <212> TYPE: PRT
257 <213> ORGANISM: Artificial Sequence
261 <220> FEATURE:
263 <223> OTHER INFORMATION: 10xHis tag
265 <400> SEOUENCE: 12
267 His His His His His His His His His
268 1
                                        10
271 <210> SEQ ID NO: 13
273 <211> LENGTH: 93
275 <212> TYPE: PRT
277 <213> ORGANISM: Escherichia coli
281 <400> SEQUENCE: 13
283 Asn Asn Gly Ala Ser Gly Ala Asp Ile Asn Asn Tyr Ala Gly Gln Ile
287 Lys Ser Ala Ile Glu Ser Lys Phe Tyr Asp Ala Ser Ser Tyr Ala Gly
291 Lys Thr Cys Thr Leu Arg Ile Lys Leu Ala Pro Asp Gly Met Leu Leu
           35
                                40
295 Asp Ile Lys Pro Glu Gly Gly Asp Pro Ala Leu Cys Gln Ala Ala Leu
299 Ala Ala Ala Lys Leu Ala Lys Ile Pro Lys Pro Pro Ser Gln Ala Val
300 65
303 Tyr Glu Val Phe Lys Asn Ala Pro Leu Asp Phe Lys Pro
304
307 <210> SEQ ID NO: 14
309 <211> LENGTH: 348
311 <212> TYPE: PRT
313 <213> ORGANISM: Artificial Sequence
317 <220> FEATURE:
319 <223> OTHER INFORMATION: TolA-BCL fusion protein
321 <400> SEQUENCE: 14
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Input Set : A:\Sequence Listing.ST25.txt
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323 Met Hi	s His H	His Hi	s His	His	Ser	Ser	Asn	Asn	Gly	Ala	Ser	Gly	Ala
324 1		5					10		_	_	_	15	
327 Asp Il		_	r Ala	Gly	Gln		Lys	Ser	Ala	Ile		Ser	Lys
328		20		_		25		m 1	0	m1	30		- 1 -
331 Phe Ty	_	Ala Se	r ser	Tyr		GLY	ьуs	Thr	Cys		ьeu	Arg	11e
332 335 Lys Le	35	Dro Ag		Mot	40	T 011	7 an	Tla	Tarc	45 B**	G1.,	C111	C1
335 Lys Le	u Ala i	PIO AS	, GIA	мес 55	пеп	пеп	Asp	116	ыуs 60	PIO	GIU	Gry	GIY
339 Asp Pr	n Δla 1	Len Cv	. Gln		Δla	T.e.11	Δla	Δla		Lvs	Len	Δla	Lvs
340 65	o niu i	bea cy	70	nια	AIG	D Cu	niu	75	1114	27.5	u	1114	80
343 Ile Pr	o Lvs 1	Pro Pr		Gln	Ala	Val	Tyr	_	Val	Phe	Lys	Asn	Ala
344	•	85					90				-	95	
347 Pro Le	u Asp 1	Phe Ly	s Pro	Gly	Gly	Gly	Ser	Gly	Ser	Leu	Val	Pro	Arg
348		100				105					110		
351 Gly Se	r Arg 1	Pro Se	r Gln	Ser		Arg	Glu	Leu	Val		Asp	Phe	Leu
352	115			_	120	_^		_	Δ	125			
355 Ser Ty	_	Leu Se	r Gln	_	Gly	Tyr	Ser	Trp		Gln	Phe	Ser	Asp
356 13	_	7 mm 7 m	- mb	135	77.	D	~ 1	~1	140	~1	Com	C1	Mot
359 Val Gl 360 145	u Giu A	ASH AL	150	GIU	Ala	PIO	GIU	155	1111	GIU	ser	GIU	160
360 145 363 Glu Th	r Pro (Ser Al		Δen	Glv	Δsn	Pro		Trn	His	T.em	Δla	
364		16		Abii	Cly	7011	170	DCI	110		Lea	175	p
367 Ser Pr	o Ala '			Ala	Thr	Ala		Ser	Ser	Ser	Leu	Asp	Ala
368		180	-			185					190	_	
371 Arg Gl	u Val	Ile Pr	o Met	Ala	Ala	Val	Lys	Gln	Ala	Leu	Arg	Glu	Ala
372 ·	195				200					205			
375 Gly As		Phe Gl	ı Leu		Tyr	Arg	Arg	Ala		Ser	Asp	Leu	Thr
376 21		T1	- ml	215	~1	m1	7.7		220	C	Dh.	~1	01 -
379 Ser Gl	n Leu l	HIS II	e Tnr 230	Pro	GIŢ	Thr	Ата	_	GIn	ser	Pne	GIU	G1n 240
380 225 383 Val Va	l Acn (Glu Le		Ara	Acn	Glv	1721	235	Trn	G137	Δra	Tle	
384	T VOII	24		Arg	лэр	GLY	250	ASII	111	Gry	nrg	255	vai
387 Ala Ph	e Phe			Glv	Ala	Leu		Val	Glu	Ser	Val		Lvs
388		260		2		265	- 4				270	-	•
391 Glu Me	t Gln '	Val Le	u Val	Ser	Arg	Ile	Ala	Ala	Trp	Met	Ala	Thr	Tyr
392	275				280					285			
395 Leu As	n Asp 1	His Le	u Glu		\mathtt{Trp}	Ile	Gln	Glu	Asn	Gly	Gly	\mathtt{Trp}	Asp
396 29		_		295					300		_	_	_
399 Thr Ph	e Val (Glu Le	_	Gly	Asn	Asn	Ala		Ala	GIu	Ser	Arg	_
400 305	- al	7 Dh	310	7	TT	Dha	T 011	315	~1	Mot	mb	7707	320
403 Gly Gl 404	n Giu A	arg Pn . 32		Arg	пр	Pne	330	1111	GIY	Mec	IIIL	335	ALG
404 407 Gly Va	l Val '			Ser	T.011	Dhe		Δτα	Lvc			333	
407 Giy va		340	- Oly	JCI	u	345	~~ -	9	273				
411 <210> SEQ ID NO: 15													
413 <211> LENGTH: 236													
415 <212>													
417 <213>	ORGANI	SM: Ar	tific	ial :	Seque	ence			•				
421 <220>	FEATUR:	Ε:											

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```
423 <223> OTHER INFORMATION: TolA-BCL fusion protein after thrombin cleavage
    425 <400> SEQUENCE: 15
    427 Gly Ser Arg Pro Ser Gln Ser Asn Arg Glu Leu Val Val Asp Phe Leu
    431 Ser Tyr Lys Leu Ser Gln Lys Gly Tyr Ser Trp Ser Gln Phe Ser Asp
                    20
                                         25
    435 Val Glu Glu Asn Arg Thr Glu Ala Pro Glu Gly Thr Glu Ser Glu Met
    439 Glu Thr Pro Ser Ala Ile Asn Gly Asn Pro Ser Trp His Leu Ala Asp
    443 Ser Pro Ala Val Asn Gly Ala Thr Ala His Ser Ser Ser Leu Asp Ala
                                                 75
    447 Arq Glu Val Ile Pro Met Ala Ala Val Lys Gln Ala Leu Arg Glu Ala
                                             90
    451 Gly Asp Glu Phe Glu Leu Arg Tyr Arg Arg Ala Phe Ser Asp Leu Thr
                    100
                                         105
    455 Ser Gln Leu His Ile Thr Pro Gly Thr Ala Tyr Gln Ser Phe Glu Gln
                115
                                     120
    459 Val Val Asn Glu Leu Phe Arg Asp Gly Val Asn Trp Gly Arg Ile Val
                                 135
            130
                                                     140
    463 Ala Phe Phe Ser Phe Gly Gly Ala Leu Cys Val Glu Ser Val Asp Lys
    467 Glu Met Gln Val Leu Val Ser Arg Ile Ala Ala Trp Met Ala Thr Tyr
                         165
                                             170
     471 Leu Asn Asp His Leu Glu Pro Trp Ile Gln Glu Asn Gly Gly Trp Asp
    472
                     180
                                         185
    475 Thr Phe Val Glu Leu Tyr Gly Asn Asn Ala Ala Glu Ser Arg Lys
                                     200
    476
                195
     479 Gly Gln Glu Arg Phe Asn Arg Trp Phe Leu Thr Gly Met Thr Val Ala
                                215
     483 Gly Val Val Leu Leu Gly Ser Leu Phe Ser Arg Lys
     484 225
                             230
     487 <210> SEQ ID NO: 16
     489 <211> LENGTH: 115
     491 <212> TYPE: PRT
    493 <213> ORGANISM: Artificial Sequence
     497 <220> FEATURE:
     499 <223> OTHER INFORMATION: Tagged TolAIII region of pTol vectors
     501 <220> FEATURE:
     503 <221> NAME/KEY: MISC FEATURE
     505 <222> LOCATION: (107)..(111)
     507 <223> OTHER INFORMATION: Xaa residues represent cleavage sites DDDDK (SEQ ID NO: 3),
LVPR
     508
               (SEQ ID NO: 4; no Xaa at position 111) or IEGR (SEQ ID NO: 5; no
              Xaa at position 111)
     513 <400> SEQUENCE: 16
     515 Met His His His His His Ser Ser Asn Asn Gly Ala Ser Gly Ala
     519 Asp Ile Asn Asn Tyr Ala Gly Gln Ile Lys Ser Ala Ile Glu Ser Lys
                     20
                                         25
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RAW SEQUENCE LISTING ERROR SUMMARY PATENT APPLICATION: US/10/501,071

DATE: 07/16/2004 TIME: 16:37:41

Input Set : A:\Sequence Listing.ST25.txt
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Please Note:

Use of n and/or Xaa have been detected in the Sequence Listing. Please review the Sequence Listing to ensure that a corresponding explanation is presented in the <220> to <223> fields of each sequence which presents at least one n or Xaa.

Seq#:16; Xaa Pos. 107,108,109,110,111

Seq#:22; Xaa Pos. 14,15 Seq#:23; Xaa Pos. 13,14 Seq#:24; Xaa Pos. 13,14 VERIFICATION SUMMARY

PATENT APPLICATION: US/10/501,071

DATE: 07/16/2004 TIME: 16:37:41

Input Set : A:\Sequence Listing.ST25.txt Output Set: N:\CRF4\07162004\J501071.raw

L:17 M:270 C: Current Application Number differs, Replaced Current Application No

L:17 M:271 C: Current Filing Date differs, Replaced Current Filing Date

L:539 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:16 after pos.:96

L:667 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:22 after pos.:0 L:701 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:23 after pos.:0 L:731 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:24 after pos.:0

STATISTICS SUMMARY

PATENT APPLICATION: US/10/501,071

DATE: 07/16/2004 TIME: 16:37:41

Input Set : A:\Sequence Listing.ST25.txt
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Application Serial Number: US/10/501,071

Alpha or Numeric or Xml: Numeric

Application Class:

Application File Date: 07-09-2004

Art Unit: PCT

Software Application: PatentIN3.1 Total Number of Sequences: 61

Total Nucleotides: 1032 Total Amino Acids: 2629

Number of Errors: 0 Number of Warnings: 4 Number of Corrections: 2

MESSAGE SUMMARY

270 C: 1 (Current Application Number differs)

271 C: 1 (Current Filing Date differs)

341 W: 4 ((46) "n" or "Xaa" used)